combination. The applicable concentration of these compound can be determined by repeated experiments when in use based on the selective solubility of HDL as an index.

In the present invention, the means for directly and/or preferentially enabling an enzymatic reaction, with respect to a component in a specific lipoprotein fraction in the reaction solution by utilizing the reaction specificity of an enzyme to a specific lipoprotein is carried out by selecting a lipoprotein lipase (LPL) and/or cholesterol esterase (CE) that preferentially act(s) on the HDL fraction. The enzyme includes commercially available LPL and CE, derived from Chromobacterium viscosum. In the case where an LDL fraction is aimed, enzymes derived from the genus Pseudomonas and the like, may be appropriately selected. The enzymes may be or do not have to be subject to various modifications as far as the enzyme activity and selectivity to the specific lipoprotein fraction are maintained. An addition amount of the enzyme is increased or decreased and controlled depending on the amount of a substrate known per se.

In the present invention, the means for directly and/or preferentially enabling an enzymatic reaction with respect to a component in a specific lipoprotein fraction in the reaction solution by utilizing the reaction selectivity of a selected nonionic surfactant to the specific lipoprotein can be identified by the HLB value of the nonionic surfactant.

In the case where an HDL fraction is aimed, one having the HLB value of 16 or more, preferably 17 or more, is selected. More

preferably, polyoxyethylene ethers having the HLB value of 17 or more are selected. The selected surfactants inhibit the enzymatic action of LPL, CE, cholesterol dehydrogenase (CDH) or the like on the LDL fraction and VLDL fraction. Specific examples thereof will be listed below. A nonionic surfactant used in the present invention can be optionally selected using HLB value as an index and is not limited to the following examples: cetyl ether (C16) (hexadecyl ether) (tradename: Nikko Chemical Co., Ltd.: BC-25TX, BC-30TX, BC-40TX), lauryl ether (C12) (dodecyl ether) (tradename: Nikko Chemical Co., Ltd.: BL-21, BL-25), oleyl ether (trade name: Nikko Chemical Co., Ltd.: BO-50), behenyl ether (C22) (tradename: Nikko Chemical Co., Ltd.: BB-30), polyoxyethylene lauryl ether (tradename: Nippon Oils & Fats Co., Ltd.: Nonion K-230), polyoxyethylene Monolaurate (tradename: Nippon Oils & Fats Co., Ltd.: Nonion S-40), polyoxyethylene ethers (tradename: Sigma: Brij 98, Brij 721, Brij 78, Brij 99) and the like.

In the case where an LDL fraction or a VLDL fraction is aimed, in particular in the case where the enzymatic reaction with a component in the LDL fraction is positively aimed, a nonionic surfactant having an HLB value of 11 to 13 is selected. Example may be made of Triton X-100, Nonion HS210, and Nonion A-10R (Nippon Oils & Fats Co., Ltd.). However, the surfactant used for assaying a component in the LDL fraction may also be optionally selected using HLB value as an index and is not limited to these thereto.

The addition amount of the above-mentioned surfactant may vary depending on the amount of lipoprotein to be assayed. The amount

shown in the case where the HDL and LDL are aimed, is about 180 μL of a reagent having a surfactant concentration of 0.01 to 10% per about 5 μL of a specimen. With such amount of the surfactant, the HDL or LDL is selectively decomposed to enable the enzymatic reaction of a component contained therein. In the case where a VLDL is aimed, the surfactant is used after adjusting a concentration to 0.05 to 20%. Thus, the VLDL is selectively decomposed to enable the enzymatic reaction of a component contained therein.

The pH of the reaction solution, in which enzymatic reaction occurs, is within such a range that lipoprotein does not cause agglutination nor makes the reaction solution cloudy and is selected taking the optimal pH of the enzyme that acts on the component in the lipoprotein. Preferably, the pH is about 6 to about 9. If the pH is about 6 or less, the lipoprotein makes the reaction solution cloudy. Although the pH around 7, at which the lipoprotein is relatively stable, may be selected for controlling the assaying conditions, the optimal pHs of the enzymes such as COD (cholesterol oxidase), CDH (cholesterol dehydrogenase), LPL, CE, and the like are also taken into consideration. Preferably, the reactions of CDH, LPL, and CE are at the pH of about 7 to about 9, and the reaction of COD is at the pH of about 6 to about 8. The reaction solution is preferably adjusted with a buffer solution and various buffer solutions usually used in biochemical reactions may be used. Examples thereof include a HEPES (2-[4-(2-hydroxyethyl)-1-piperazinyl] ethanesulfonic acid) buffer solution, a PIPES (piperazine-1,4'-bis(2-ethanesulfonic acid))